

http://www.cim.uh.cu/rim/

revista de investigaciones marinas	

## ARTICULO ORIGINAL

# ADVANCES IN SPOTTED ROSE SNAPPER (*LUTJANUS GUTTATUS*, STEINDACHNER, 1869) JUVENILES PRODUCTION

Avances en la producción de juveniles del pargo flamenco (*Lutjanus guttatus*, Steindachner, 1869)

Leonardo Ibarra-Castro<sup>1,2\*</sup>, Martín Omar Ochoa-Bojórquez<sup>1</sup>, Juan Luis Sánchez-Tellez<sup>1</sup>, Angel Humberto Rojo-Cebreros<sup>1</sup> and Luis Álvarez-Lajonchère<sup>3</sup>

- <sup>1</sup> Centro de Investigación en Alimentación y Desarrollo (CIAD), A. C., Unidad Mazatlán, Avenida Sábalo Cerritos S/N, Mazatlán, C.P. 82010, A.P. 711, Sinaloa, México.
- <sup>2</sup> University of Florida Whitney Laboratory for Marine Bioscience; 9505 Ocean shore Blvd, St Augustine, Florida, USA; Phone: +1 (904)2018421.
- <sup>3</sup> Gr Piscimar, Calle 41 No. 886, Nuevo Vedado, Plaza, La Habana, CP 10600, Cuba.
- \* l.ibarracastro@whitney.ufl. edu

Recibido: 04-08-2020

Aceptado: 04-09-2020

#### ABSTRACT

This study describes the advances in spotted rose snapper Lutjanus guttatus juveniles production. The broodstock (three groups, each of ten females, mean  $\pm$  SD: 0.91  $\pm$  0.2 kg and 20 males 1.2  $\pm$  0.3 kg) with two years, were acclimated for 1.5 years in three communal cylindrical fiberglass tanks (18-m<sup>3</sup>). Fertilized eggs were obtained during natural spawning seasons. Floating eggs (average  $90 \pm 2\%$ ) were collected between 12 to 14 h after spawning. An average of 97% of them were transparent with live embryos. For the larval rearing process, initial stocking densities were  $196 \pm 70$  embryos/l, incubated in twelve  $6 - m^3$  cylindrical fiberglass tanks with 3-m<sup>3</sup> of water. The incubation period was 20 h at 26-28 °C. Hatching and survival at 48-h post-hatch (hph) were  $90 \pm$ 2% and  $55 \pm 10\%$ , respectively, with an average of  $281,258 \pm 92,202$  48-h larvae per tank and  $105 \pm 33$  larvae/l at first feeding. A total of 338.812 juveniles with 45 days post-hatched (dph) were harvested, at an average of  $28,234 \pm 11,890$  per larval rearing tank (2.4 $\pm 1.0/l$ ), and 180,000 juveniles (BW: Body weight of 5-8 g) from the nursery tanks with 90 dph. Average survival from first feeding to end of 45 dph rearing period was  $12 \pm 9\%$ . The average harvest weight was  $0.47 \pm 0.10$  g BW and the average final biomass per larval tank was 13 kg. An efficiency index at 45 days culture was 10 / 48-h larvae/juvenile, while at 90 dph the efficiency index was 19 / 48-h larvae/juvenile. The result showed that changes to the previous culture protocols of the CIAD-Mazatlan fish plant, ensure a higher production of juveniles.

**KEY WORDS**: snapper, *Lutjanus guttatus*, massive production, larval and juvenile rearing

#### RESUMEN

El presente estudio describe los avances en la producción de juveniles del pargo flamenco Lutianus guttatus. Los reproductores (tres grupos, cada uno de 10 hembras, media  $\pm$  SD:  $0.91 \pm 0.2$  kg y 20 machos  $1.2 \pm 0.3$  kg) con dos años, fueron aclimatados por 1.5 años en tres tangues comunales cilíndricos de fibra de vidrio (18-m<sup>3</sup>). Los huevos fertilizados fueron obtenidos durante las temporadas naturales de desove. Los huevos flotantes (promedio de  $90 \pm 2\%$ ) se colectaron entre 12 a 14 horas post-desove. En promedio, el 97% de ellos fueron transparentes, con un embrión vivo. La densidad inicial de siembra para la cría larval fue de  $196 \pm 70$  embriones/l, incubados en doce tanques cilíndricos de 6-m<sup>3</sup> con 3-m<sup>3</sup> de agua. El periodo de incubación fue de 20 h a 26-28 °C. Los porcentajes de eclosión y de supervivencia a las 48- horas post-eclosión (hpe) fueron  $90 \pm 2\%$  y  $55 \pm 10\%$ , respectivamente, con un promedio de larvas de 48-hpe de 281,258  $\pm$  92,202 por tangue y 105  $\pm$  33/l. Se cosechó un total de 338,812 juveniles con 45 días después de la eclosión (dpe) a una densidad promedio de  $28,234 \pm 11,890$  por tangue (2.4 ± 1.0/l). En el alevinaje se cosecharon 180,000 juveniles de 90 dpe (peso del cuerpo: 5-8 g). La supervivencia desde la primera alimentación hasta los 45 dpe fue de  $12 \pm 9$  %, con un peso medio de  $0.47 \pm 0.10$  g y una biomasa final por tanque de 13 kg. El índice de eficiencia a los 45 días de cría fue de 10 larvas de 48 hpe por juvenil producido, mientras que a los 90 dpe el índice fue de 19 larvas de 48 hpe por juvenil producido. Los resultados mostraron que los cambios a los previos protocolos de cultivo de la planta de peces CIAD-Mazatlán, aseguran una mayor producción de juveniles.

PALABRAS CLAVE: pargo, *Lutjanus guttatus*, producción masiva, cría de larvas y juveniles

#### INTRODUCTION

The snappers (family Lutjanidae) are highly valued for human consumption, with a high and unsatisfied market demand around the world. Snapper fisheries are considered overexploited in many areas (Davis et al., 2000; Amorim et al., 2019), and their culture production is still limited (FAO, 2019). Red mangrove snapper *Lutjanus argentimaculatus* and John's snapper *Lutjanus johnii* are the most cultured snapper species, in the Indo-Pacific region (FAO, 2019).

In general, snapper aquaculture has depended on unsustainable and unpredictable capture of wild juveniles in coastal waters to stock most grow-out facilities (Davis et al., 2000; Surtida and Buendia, 2002). Although, there are some protocols to produce juveniles in pilot hatchery conditions, e.g. mutton snapper Lutianus analis (Watanabe et al., 1998; Benetti et al., 2002), red mangrove snapper Lutianus argentimaculatus (Duray et al., 1996; Leu et al., 2003), red snapper Lutjanus campechanus (Ogle and Lotz, 2006) and yellowtail snapper Ocyurus chrysurus (Turano et al., 2000; Gutierrez-Sigeros et al., 2018), the juveniles mass production is still inconsistent in many facilities.

The main requirement for commercial aquaculture is to establish a mass juvenile production protocol (Tucker, 1998). Therefore, is crucial to develop new procedures for stable, reliable, and cost-effective mass production at a pilot-commercial scale. In this context, spotted rose snapper L. guttatus is an emerging species with commercial importance in Mexico and other Latin American countries. In consequence, research is being carried out in Mexico and Costa Rica to mass-produce this species for commercial purposes. The commercial culture has been established in Costa Rica (Sardenberg et al., 2014), but a description of constant juvenile mass production procedures, has not been reported yet, except the preliminary work by Alvarez-Lajonchere et al. (2012).

Research with spotted rose snapper at the Research Centre for Food and Development (CIAD) in Mazatlan (Mexico), started in 2003 with wild matured breeders recently caught (Ibarra-Castro and Duncan, 2007), followed by the experimental production of juveniles (Abdo de la Parra et al., 2010). The first larval rearing at pilot-scale produced 22 000 juveniles and were harvested from a single trial using naturally spawned eggs from a captive broodstock (Ibarra-Castro and Alvarez-Lajonchère, 2009, 2011; Alvarez-Lajonchère et al., 2012). Consequently, the purpose of this study is to describe and examine the results of the follow-up research on juvenile mass production procedures on a pilot-scale at CIAD-Mazatlan fish plant, modifying the traditional protocol to apply it at a commercial scale.

## **MATERIAL AND METHODS**

#### **EGG PRODUCTION**

Three groups of ten females  $(0.91 \pm 0.2 \text{ kg})$ body weight (BW) and 20 males  $(1.2 \pm 0.3 \text{ kg})$  BW) each, after their first spawning season with two years, were acclimatized for one and a half years in three communal maturation spawning cylindrical fiberglass tanks (3.5-m diameter by 2.0 m deep, 18-m<sup>3</sup>). During the acclimation period, no samplings were achieved to estimate sexual development or growth. The young fish (2+ years of age) were held in a flow-through system (8 tank volumes/day), strong aeration and covered by shade cloth (70%).

For broodstock feeding and management, as well as spawned eggs handling, the procedure described by Ibarra-Castro and Alvarez-Lajonchère (2011) was followed. Briefly, each morning after 12 to 14 h after fertilization, eggs were removed from collector at somitogenesis stage. The number of floating and sinking eggs were estimated volumetrically using a 500-ml graduated cylinder, based on the equation E = -18.648 D + 16645; P = 0.05, r<sup>2</sup> = 0.9013 where E is the total number of eggs in 1 ml, and D is the average egg diameter of 50 eggs (Ibarra-Castro and Alvarez-Lajonchère 2011). To estimate the viability of floating eggs, a sample of >120 eggs was used and the percent of floating eggs with live embryos was recorded. Egg and oil droplet diameters were measured using a composed microscope Olympus BX41<sup>®</sup> with a calibrated ocular micrometer ( $\pm$  10µm) at 4X. Before stocking the eggs in the larval rearing tanks, they were rinsed in fresh water for 3 minutes followed by immersion for 10 minutes in presence of 2 ppm chlorine dioxide (Pulih Purifica<sup>®</sup>) solution as a prophylactic treatment to prevent infection.

## LARVAL AND JUVENILE REARING ENVIRONMENT MANAGEMENT

Incubation and larval rearing were carried out in 6-m<sup>3</sup> cylindrical fiberglass tanks with black walls and white bottoms. The initial working volume was 3 m<sup>3</sup> of water, and after the first week, it was increased to  $4m^3$ , then to  $5m^3$  and finally to  $6-m^3$  during the second week (Fig 1). Hatching percentages for each tank were determined by incubating 50 eggs with live embryos in three 1-L beakers, recording the number of newly hatched larvae the next morning. The same procedure was used to estimate the viable percentages of larval survival at 48-h post-hatch (hph), defined as a normal larval (straight, with open mouth, and pigmented eves).

Seawater was pumped from a sub-sand system (Alvarez-Lajonchère et al., 2007) to



Fig. 1. Water quality and environmental management protocol during incubation, larval, and juvenile rearing of spotted rose snapper, *Lutjanus guttatus*.

a 50 m<sup>3</sup> reservoir and from it into two elevated 10 m<sup>3</sup> tanks and passed through two parallel pressurized sand filters, followed by multi-cartridge filtration (16  $\mu$ m relative retention). Before supplying the water to the larval rearing section, this was recirculated through a heat-pump (Air Energy Model AE1000) to control the temperature at 26 °C. After that, the water was filtered again through a multi-cartridge 10-5-1  $\mu$ m retention unit and finally passed through four UV-lamps (60 mJ cm<sup>-2</sup> each). Through the egg incubation period and larval rearing process, a protocol for the water quality and environmental management was established (Fig 1). Control practices, cleaning, feeding, and behavioral observations, were made following Alvarez-Lajonchère et al. (2002).

#### LIVE FEED PRODUCTION, LARVAL AND JUVENILE FEEDING REGIME

Rotifers *Brachionus sp* culture procedures in the present study were described in detail by Rojo-Cebreros et al. (2017). The larvae were fed using rotifers from 2 to 20 days post-hatched (dph) (Fig 2). Harvested rotifers were enriched with DHA Protein Selco<sup>®</sup> (INVE Aquaculture Inc., Mazatlan Mexico) at 150 mg/l for 18 h at a density of 450 rotifers/ml.

Larvae were fed manually. Daily work started at 06:00 am by counting the remaining rotifers in the tank and repeating



Fig. 2. Feeding regimen during larval and juvenile rearing of spotted rose snapper, Lutjanus guttatus.

this procedure every four hours. Rotifers densities in the rearing tanks were maintained at 5-10/ml for early larvae up day 5 dph and increased to 15-20/ml from 6 dph to 20 dph (Fig 2). Artemia was supplied from 15 dph to 30 dph, the first 5 days as newly hatched nauplii, and the between 18 dph to 30 dph enriched metanauplii (A1 DHA Selco<sup>®</sup>, INVE Aquaculture Inc., Mazatlan Mexico), following manufacturer protocols. Each day Artemia was supplied on demand after counting the remaining individuals in the tank (every four hours). The Artemia nauplii densities in the rearing tank were 0.5 to 1/ml, while Artemia metanauplii were increased from 2 to 5/ml, throughout the culturing period. To maintain the quality of food during the day, Artemia nauplii and metanauplii were stored at 8°C. Weaning was initiated on 12<sup>th</sup> dph (Otohime<sup>®</sup>, from Proaqua, Mexico) supplied twelve times per day (Fig 2). The small juveniles were fed with a mix from 0.8 mm to 3 mm, from day 30 to 90 dph using two different brands of marine fishmeal, Otohime<sup>®</sup> and SKRETTING<sup>®</sup> from Proaqua, Mexico.

#### LARVAL REARING, NURSERY, AND SAMPLING OF LARVA AND JUVENILES

A traditional rearing cycle was repeated in twelve  $6\text{-m}^3$  tanks using naturally spawned previously described. The rearing protocol was similar to the one described by Alvarez-Lajonchère et al. (2012), but applied with slight changes: larger tanks ( $6 \text{ m}^3 \text{ vs } 3 \text{ m}^3$ ), without the use of copepods, increasing the water flow, a stronger water treatment and major control physicochemical variables during the culture. After prophylactic treatment, eggs were incubated at  $3 \text{ m}^3$  as initial working volume and, increased gradually to  $6\text{-m}^3$ . Larvae were grown to 45 dph in the same tanks, after that, they were harvested and transferred to eighteen 5-m<sup>3</sup> cylindrical fiberglass tanks for a nursery stage until 90 dph without size-grading.

For general observations, the larvae and juveniles were immobilized with 0.15 ml/l of 2-phenoxyethanol (Sigma-Aldrich, Inc, Toluca, Mexico) and observed with a compound microscope Olympus BX41<sup>®</sup> or using a stereoscope Olympus SZX16<sup>®</sup>. Juvenile growth was monitored every four to five days by measuring the total length (TL) of 25 individuals sampled at random from each tank. Larvae were sampled after concentrating them with a net. To take the larvae, a plastic 1-L beaker was used during the first two weeks. Hand nets were used afterward: briefly, in the 5-m<sup>3</sup> cylindrical fiberglass tanks, the water level of the tank was lowered to decrease larvae sampling stress. To estimate the numbers of harvested larvae, a gravimetric method was used following Schipp et al. (2007). Water quality parameters during egg incubation, larval and juvenile rearing were temperature  $26 \pm 2.0$  °C, salinity  $35 \pm 1.0$  g/l, dissolved oxygen 6.1  $\pm$ 0.6 mg/l (saturation:  $88.8 \pm 9\%$ ), pH 8 ± 0.2 and NH<sub>3</sub> lower than 0.05 mg/l.

## RESULTS

#### **E**GG PRODUCTION AND QUALITY

During the whole production season, a total of 272 spawns were registered, with a total of 228.15 million eggs spawned, 92.9 % were floating eggs, and an average viability of 95 %. The average diameter in floating eggs stocked in the rearing tanks was  $760 \pm 33 \mu m$  and the average in oil droplet diameter was  $123 \pm 10 \mu m$ .

From whole eggs spawned, a total of 7,062,039 eggs were stoked in twelve

different culture tanks. An average of  $588,503 \pm 209,477$  of embryos was stoked per tank, with an initial volume of 3 m<sup>3</sup>, at densities of  $196 \pm 70$  embryos/l. The incubation period was 20 h at 26-28 ° C with average hatching of  $88 \pm 2\%$ . The average larvae survival at first feeding (48-h) was  $55 \pm 10\%$ , with an average of  $281,258 \pm 92,202$  48-h larvae per tank and  $105 \pm 33$  larvae/l at first feeding, several more times than in previous rearing trials.

## **P**RODUCTION AND SURVIVAL OF LARVAE AND JUVENILES

At ambient conditions of  $26 \pm 1.0$  °C and salinity  $35 \pm 1.0$  g/l, the larvae opened their mouths at the end of day 2 post-hatching, and the first feeding started when the yolk sac was nearly absorbed. Rotifers were observed in the larval guts in the early morning of the third dph, 8 to 12 h after their first supply. The *Artemia* nauplii and enriched metanauplii were observed in the larvae digestive tract on the first day that they were supplied. Cofeeding involving rotifers and *Artemia*, together with artificial food, was achieved on the 15<sup>th</sup> dph (Fig 2).

A total of 338,812 juveniles were harvested, at an average of  $28,234 \pm 11,890$  per rearing tank, and an average survival from the first feeding to end of 45 dph rearing period was  $12 \pm 9$  %. To produce those juveniles, it was required to stock 10 first feeding larvae per juvenile. The average harvest weight was  $0.47 \pm 0.10$  g BW at the end of the 45-d rearing. Finally, the average final harvest biomass per tank was 13 kg BW/tank. In nursery from 5-m<sup>3</sup> tanks, 180,000 juveniles were harvested with a BW of 5-8 g, which represented an efficiency index of 19 / 48-h larvae/ juvenile.

## **G**ENERAL OBSERVATIONS ABOUT MORPHOLOGICAL DEVELOPMENT AND GROWTH IN LARVAE AND JUVENILES

Newly hatched larvae were transparent and presented a broad yolk sac that extended forward to snout and in front, it was observed the oil droplet below the head, there were no other structures detected. Total length (TL) larval size at hatch varied between 1.18 to 1.4 mm. At the first feeding step, 54 to 72 hph, larvae showed morphology changes like eye pigmentation, mouth opening, functional jaw, esophagus, and three intestine division (anterior-midposterior). Organs like the liver, pancreas, gallbladder, and swim bladder were observed. To cover the energy demands before first feeding, the spotted rose snapper, L. guttatus larvae consumed the total volk reserve, and oil droplet between 38 h and 56 hph, and the range of larvae size at first feeding 48 hph was between 2.2 to 2.7 mm. Also, larvae growth slowed down during the first three weeks of culture (TL 2.6 to 6.02 mm) (Fig 3). Between the fourth and fifth weeks in larval rearing, the larvae grew up from TL of 7 to 11 mm (Fig 3). In the nursery step at the end of the sixth week, the average body weight of juveniles was 7.34 g (Fig 4).

## DISCUSSION

Total eggs spawned were higher than in other snappers with long reproduction periods as reviewed by Alvarez-Lajonchère et al. (2012). Variables such as percentages of floating eggs with live embryos at collection time and hatching showed high values ( $\geq$  90%), like those obtained by Ibarra-Castro and Alvarez-Lajonchère (2011), but at 48-h post-hatch, survival percentages of the present study were lower (55 %). These low percentages of larval survival at the



**Fig. 3.** Growth in total length (mm) curve of spotted rose snapper, *Lutjanus guttatus*, larvae at water temperatures of  $26 \pm 1.0 \text{ °C}$  and salinity of  $35 \pm 1.0 \text{ g L}^{-1}$ . Each point represents the average  $\pm$  standard error of the average of 25 fish sampled.

first feeding step were not a limitation to start the process of juvenile production, due to the great number of larvae produced. The reduction in the availability of larvae at first feeding could be due to the broodstock age (Carrillo et al., 2000), which affected the quality of the eggs during the first reproduction seasons as it has been reported for other species (Carrillo et al., 2000). Clearly defining a good quality in eggs for snapper cultures has been difficult, as reported by Bardon-Albaret and Saillant (2017).

During the present work, water quality variables were considered suitable for the species. However, dissolved oxygen levels in broodstock tanks (5 mg/l) were lower than those characteristics of the species spawning grounds (6 mg/l). To improve the maintenance, the establishment of a recirculation system and/or using packed columns could improve the oxygen level (Timmons and Ebeling, 2010). Stabilizing environmental conditions could have a direct effect on the quality of the spawned eggs and larval survival.

Larval rearing survival percentages during the rearing period were stabilized in the present study, compared with those of previous experimental research trials (Abdo de la Parra et al., 2010), the first pilot-scale trial (Alvarez-Lajonchère et al., 2012) and earlier pre-commercial trials at CIAD-Mazatlan fish plant (Ibarra-Castro et al., not published), although in the present work the juveniles were not size graded. The average survivals achieved for this species in Costa Rica (Boza-Abarca et

al., 2008; Herrera-Ulloa et al., 2010) was also lower than in the present work. The increase in survival during larval culture in pilot-scale condition could be influenced by better water quality and stability of the parameters, together with strict control of feeding with live prey, and formulated feed supplied earlier. Best survival results were reported by Schipp et al. (2001) with *L. johnii*, using a mesocosmos technique, followed by Leu et al. (2003) with *L. argentimaculatus* (Table 1).

As in other marine fish larvae, high mortalities occur during the larval rearing cycle. First, mortality was observed after the exogenous first feeding phase, which may be due to the few reserves of yolk and oil droplet, which is characteristic of this group of species (Davis et al., 2000). Besides, in the first rearing week, the larvae spend energy on developing all the structures and organs necessary to feed, which can contribute to more drastic mortality. The second mortality period was observed through the metamorphosis phase (final inflation of the swim bladder, dorsal and pelvic fins appearance as well as torsion of the mid intestine). This mortality was probably influenced by poor development of internal organs, hyper swim bladder inflation, poor nutrition, and especially by cannibalism. The same periods of mortality were reported for this species by Boza-Abarca et al. (2008) and Alvarez-Lajonchère et al. (2012), as well as for other snapper species (Tucker, 1998). These two critical periods are also frequent in other marine fish species (Kraul et al., 1993;



**Fig. 4.** Growth curve in weight (g) of spotted rose snapper, *Lutjanus guttatus*, partially metamorphosed juveniles and juveniles at water temperatures of 26 to 31 °C and salinity of 33 to 35 g/l. Each point represents the average  $\pm$  standard error of the average of 25 fish sampled.

present work (modified from	m Alvarez-Lajo	onchère et al., 20	12).	6					
Species	Rearing tanks (m <sup>3</sup> )	Stocking den- sity (larvae/l)	Duration (days)	Survival (%)	Final den- sity (No./I)	W (g)	Biomass (kg/m³)	Juveniles (x 10 <sup>3</sup> )	Authority
Mangrove red snapper, Lutjanus argentimaculatus	3 x 9 t	30	55	12	0.6	ca. 0.54 <sup>b</sup>	ca. 0.3°	10.2	Duray et al. (1996)
Spotted rose snapper, Lutjanus guttatus	6 x 2 t x 9 cycles	33	55-60	2	0.66	I	ł	20	Herrera-Ulloa et al. (2010)
John's snapper, <i>Lutjanus johnii</i>	40 x 2 t	5	35-40	24-35	0.5-0.7			56	Schipp et al. (2001)
Mutton snapper, Lutjanus analis	30 x 1 t	8,6	38	14.3	1.2	0.31±0.02	0.38	36.9	Watanabe et al. (1998)
Spotted rose snapper, Lutjanus guttatus	3 x 6 t	15	60	12.1±1.1	1.3±0.2	2.4±0.04	2.8	22.6	Alvarez-Lajonchere et al. (2012)
Mangrove red snapper, Lutjanus argentimaculatus	4 x 6 t	9.9±4	50	21.1±7	2.0±1.0	ca. 2.1 <sup>b</sup>	ca. 4.2°	48.9	Leu et al. (2003)
Spotted rose snapper, Lutjanus guttatus	6 x 12 t	105±33	45	12±9	2.4±1.0	0.47±0.10	2.2	338.8	Present study
t = Tank number b = Estimated considerir c = Calculated	1g TL = FL x	t 1.018 and W =	= 0.0280 F	L <sup>2.844</sup> (ww	w.fishbase.o	rg)			

SPOTTED ROSE SNAPPER (LUTJANUS GUTTATUS) JUVENILES PRODUCTION

Ibarra-Castro • Ochoa-Bojórquez • Sánchez-Téllez • Rojo-Cebreros • Álvarez-Lajonchère

10

**REVISTA DE INVESTIGACIONES MARINAS** RNPS: 2096 • ISSN: 1991-6086 • VOL. 40 • No. 2 • JULIO-DICIEMBRE • 2020 • pp. 1-14

Tucker, 1998). In contrast, Watanabe et al. (1998) and Leu et al. (2003) did not report a critical period during the first two weeks in mutton snapper and red mangrove snapper, respectively.

Mortality during first feeding may be reduced with the use of smaller rotifers 80 um (neonates) or even a smaller species, as Proales similis (Hagiwara et al., 2014). During the first four to five days of larval rearing, it is essential to provide small size rotifers. Other small foods for early larvae are copepod nauplii and copepodites, which have the size and nutritional value required by the larvae of many marine fishes (Tucker, 1998). The use of copepods as the first food has been tested in other snappers (Singhagraiwan and Doi, 1993; Schipp et al., 2001; Leu et al., 2003; Ogle and Lotz, 2006) with good results. However, copepod production has logistic difficulties that lower the feasibility of larval mass production, considering challenges of generating cultured copepods at a large scale, therefore copepods were not supplied during the present trials.

The second critical period of mortality is a complicated stage of metamorphosis on which important morphological and physiological changes take place, as explained before. At this time, vital changes take place in the morphology and physiology of the larvae. Also, the differential growth present leads to size hierarchies within the culture, which increased cannibalism, starting during the second week in the present species in previous rearing cycles, much earlier than in other snapper species, as reviewed by Alvarez-Lajonchère et al. (2012).

As discussed previously by Alvarez-Lajonchère et al. (2012), the growth of the larvae was slow during the rotifers feeding

period and increased after Artemia metanauplii were supplied. Growth in the present study was much slower than in other snappers, but similar to mutton snapper (Watanabe et al., 1998) and other reports of spotted rose snapper (Boza-Abarca et al., 2008; Alvarez-Lajonchère et al., 2012). In addition, final densities were high, only slightly smaller than those reported by Leu et al. (2003) with L. argentimaculatus, and the present study (Table 1). As a consequence of related to this parameter, final biomasses also showed higher values reported by Leu et al. (2003) with L. argentimaculatus, Alvarez-Lajonchere et al. (2012) and the present report with L. guttatus (Table 1). The results obtained, especially juvenile production and efficiency could be improved by the following protocol changes: a) a significant increase in the initial stocking density; b) modifications to the feeding regime, as a continuous live food supplied, with an extended supply of rotifers and microalgae (to 25 dph), and earlier supply of *Artemia* and micro-diets; c) a shorter larval rearing period; d) an intensive nursery stage in smaller tanks with low water depth and strong water current; e) regular size grading. Similar rearing techniques have been successfully applied to reduce cannibalism in other finfish species (Hecht and Piennar, 1993; Ostrowski et al., 1996; Schipp et al., 2007).

### CONCLUSION

The research conducted showed that the procedures and management broodstock assured production of eggs and larvae. Also, the changes in the previous rearing protocol was successful for larval and juvenile production and that it is feasible to apply it at a commercial scale. Our recommendation for future researches is a combination

of changes suggested, together with a financial analysis to evaluate the feasibility when implementing the rearing protocol to a commercial scale. However, five rearing cycles with the present protocol could reach a minimum of one million juveniles in a year-round production. This number of juveniles was considered as a commercial scale by Kuronuma and Fukusho (1984).

## ACKNOWLEDGMENTS

The authors are thankful to the staff and students of the Fish Plant at CIAD Mazatlan Unit for their essential assistance. This study was supported by grants of the National Aquaculture and Fishery Commission (CONAPESCA) to L. Ibarra-Castro.

## REFERENCES

- Abdo-de la Parra, M. I., Rodríguez-Ibarra, L. E., Campillo-Martínez, F., Velasco-Blanco, G., García-Aguilar, N., Alvarez-Lajonchère, L. & Voltolina, D. (2010). Efecto de la densidad de siembra sobre el crecimiento y supervivencia larval del pargo lunarejo *Lutjanus guttatus. Rev. Biol. Mar. Oceanogr., 45*, 141-146.
- ALVAREZ-LAJONCHÈRE, L., ABDO DE LA PARRA,
  M. I., RODRÍGUEZ-IBARRA, L. E., VELASCO
  BLANCO, G., PUELLO CRUZ, A., GONZÁLEZ
  RODRÍGUEZ, B., ... N., IBARRA SOTO, A.
  (2012). Scale-up of spotted rose snapper, *Lutjanus guttatus*, larval rearing at
  Mazatlan, Mexico. J. World Aquacult.
  Soc., 43, 411-422.
- ALVAREZ-LAJONCHÈRE, L., CERQUEIRA, V. R., SILVA, I, ARAÚJO, D. J & DOS REIS, M. (2002). Mass production of juveniles of the fat snook *Centropomus parallelus* in Brazil. J. World Aquacult. Soc., 33, 506-516.

- ALVAREZ-LAJONCHÈRE, L., REINA-CÁÑEZ, M. A., CAMACHO-HERNÁNDEZ, M. A. & KRAUL, S. (2007). Design of a pilot-scale tropical marine finfish hatchery for a research center at Mazatlan, Mexico. *Aquacult. Eng.*, *36*, 81-96.
- AMORIM, P., SOUSA, P., JARDIM, E. & MENE-ZES, G. M. (2019). Sustainability status of data-limited fisheries: global challenges for snapper and grouper. *Front. Mar. Sc.*, 6(654), 1-17. http://www.frontiersin. org. Doi: 10.3389/fmars.2019.00654.
- SURTIDA, M. B., & BUENDIA, R. Y. (2002). The emerging red snapper culture. *Asian Aquacult.*, 24 (4), 23-24, 28. http:// repository.seafdec.org.ph.
- BARDON-ALBARET A. & SAILLANT, E. (2017). Egg quality traits and predictors of embryo and fry viability in red snapper Lutjanus campechanus. Aquacult. Rep., 7, 48-56.
- BENETTI D., MATERA, J., STEVENS O. M., ALARCON J. F., HAEWL, F., ROTMAN, F., ... ALDRIDGE, L. (2002). Growth, survival, and feed conversion rates of hatcheryreared mutton snapper *Lutjanus analis* cultured in floating net cages. *J. World Aquacult. Soc.*, 33, 349-357.
- BOZA-ABARCA, J, CALVO-VARGAS, E., SOLIS-ORTIZ, N. & KOMEN, J. (2008). Induced spawning and larval rearing of spotted rose snapper, *Lutjanus guttatus*, at the Marine Biology Station, Puntarenas, Costa Rica. *Cien. Mar.*, 34, 239-252.
- CARRILLO, M., ZANUY, S., OYEN, F., CERDÁ, J. NAVAS, J. M. & RAMOS, J. (2000). Some criteria of the quality of the progeny as indicators of physiological broodstock fitness. In *Recent advances in Mediterranean finfish species diversification. Proceedings of the seminar of the CIHEAM Network on technology of aquaculture in the Mediterranean*

*(TECAM)* (pp. 63-73). Jointly organized by CHEAM and FAO, Zaragoza (Spain), 24-28 may 1999. Cahiers, 47.

- DAVIS, D. A., BOOTES, K. L. & ARNOLD, C.
  R. (2000). Snapper (Family Lutjanidae) culture. En R. R. Stickney (Ed.), *Encyclopedia of aquaculture* (pp. 884-889).
  John Wiley and Sons, Inc., New York.
- DURAY M. N., APLAZAN, L. G. & ESTUDILLO, C. B. (1996). Improved hatchery rearing of red mangrove snapper, *Lutjanus* argentimaculatus, in large tanks with small rotifer (*Brachionus plicatilis*) and *Artemia. Isr. J. Aquacult./Bamidgeh*, 48, 123-132.
- FAO (2019). FISHSTAT Plus: Universal software for fishery statistical time series. Version 20191.0.
- GUTIERREZ-SIGEROS, I., IBARRA-CASTRO, L. ALVAREZ-LAJONCHÈRE, L. & SANCHEZ-ZAMORA, A. (2018). Natural spawning and scaling-up of yellowtail snapper (Ocyurus chrysurus): Larval rearing for the mass production of juveniles. Aquaculture, 491, 252-257.
- HAGIWARA, A., WULLUR, S., MARCIAL, H. S., HIRAI, N. & SAKAKURA, Y. (2014). Euryhaline rotifer *Proales similis* as initial live food for rearing fish with small mouths. *Aquaculture*, 432, 470-474.
- HECHT, T. & PIENAAR, A. G. (1993). A review of cannibalism and its implications in fish larviculture. J. World Aquacult. Soc., 24, 246-261.
- HERRERA-ULLOA, A., CHACÓN-GUZMÁN, J., ZÚÑIGA-CALERO, G. & JIMÉNEZ-MON-TEALEGRE, R. (2010). Spotted rose snapper (*Lutjanus guttatus*) aquaculture research and development as socio-economic alternative for Costa Rican fisheries communities. World Aquacult., 41, 20-22.
- IBARRA-CASTRO, L. & ALVAREZ-LAJONCHÈRE, L. (2009). An improved induced-spawning

protocol for spotted rose snapper Lutjanus guttatus. Isr. J. Aquacult./ Bamidgeh, 61, 121-133.

- IBARRA-CASTRO, L. & ALVAREZ-LAJONCHERE, L. (2011). GnRHa induced multiple spawns and volition spawning of captive spotted rose snapper (*Lutjanus guttatus*) at Mazatlan, Mexico. J. World Aquacult. Soc., 42, 564-574.
- IBARRA-CASTRO, L. & DUNCAN, N. J. (2007). GnRHa-induced spawning of wildcaught spotted rose snapper *Lutjanus guttatus*. *Aquaculture*, 272, 737-746.
- KRAUL, S., BRITTAIN, K., CANTRELL, R., NAGAO, T., AKO, H., OGASAWARA, A. & KITAGAWA, H. (1993). Nutritional factors affecting stress resistance in the larval mahimahi, *Coryphaena hippurus*. J. World Aquacult. Soc., 24, 186-193.
- KURONUMA, K. & FUKUSHO, K. (1984). Rearing of marine fish larvae in Japan. Ottawa, IDRC-TS473.
- LEU, M.-Y., CHEN, I.-H. & FANG, L.-S. (2003). Natural spawning and rearing of mangrove red snapper, *Lutjanus argentimaculatus*, larvae in captivity. *Isr. J. Aquacult./Bamidgeh*, 55, 22-30.
- OGLE, J. T. & LOTZ, J. M. (2006). Characterization of an experimental indoor larval production system for red snapper. *N. Am. J. Aquacult.*, *68*, 86-91.
- OSTROWSKI, A. C., IWAI, T., MONAHAN, S., UNGER, S., DAGDAGAN, D., MURAKAWA, P., ... PIGAO, C. (1996). Nursery production technology for Pacific threadfin (*Polydactylos sexfilis*). Aquaculture, 139, 19-39.
- ROJO-CEBREROS, A. H., IBARRA-CASTRO, L., GUERRERO-CARLOCK, E., SÁNCHEZ-TÉLLEZ, J. L. & ALVAREZ-LAJONCHÈRE, L. (2017). Producción piloto del rotífero *Brachionus* sp. bajo diferentes sistemas de cultivo. *Rev. Biol. Mar. Oceanogr.*, 52, 539-549.

- SARDENBERG, B., BENETTI, D. D., LARA, C., LUTZ, R. P., BUCHALLA, Y., SOLIGO, T., ... SUAREZ, J. (2014). Development and implementation of vertically integrated aquaculture of Pacific red snapper Lutjanus guttatus in Costa Rica. III Aquaculture Investment Workshop 2014, La Paz, Baja California Sur, México.
- SCHIPP, G. R., BOSMANS, J. M. P. & GORE, D.
  J. (2001). A semi-intensive larval rearing system for tropical marine fish. En
  C. I. Hendry, G. Van Stappen, M. Wille, P. Sorgeloos, (Eds.), Larvi' 01-Fish
  & Shellfish Larviculture Symposium (pp- 536-539). European Aquaculture Society, Special Publication, Oostende, Belgium.
- SCHIPP, G., BOSMANS, J. & HUMPHREY, J. (2007). Northern Territory barramundi farming handbook. Northern Territory Department of Primary Industry, Fisheries and Mines. Technical Publication.

- SINGHAGRAIWAN, T. & DOI, M. (1993). Biology and culture of the red snapper, Lutjanus argentimaculatus. The Eastern Marine Fisheries Development Center and Japan International Cooperation Agency, Thailand.
- TIMMONS, B. M. & EBELING J. M. (2010). Recirculating aquaculture. NRCA, New York.
- TUCKER, J. W., JR. (1998). *Marine fish culture*. Kluwer Academic Publishers, Boston.
- TURANO, M. J., DAVIS, D. A. & ARNOLD, C. R. (2000). Observations and technologies for maturation, spawning, and larval rearing of the yellowfin snapper Ocyurus chrysurus. J. World Aquacult. Soc., 31, 59-68.
- WATANABE, W. O., ELLIS, E. P., ELLIS, S. C., CHAVES, J., MANFREDI, C., HAGOOD, R. W., ... ARNESEN, S. (1998). Artificial propagation of mutton snapper *Lutjanus analis*: a new candidate marine fish species for aquaculture. J. World Aquac. Soc., 29, 176-187.

#### **C**OMO CITAR ESTE ARTÍCULO

Ibarra-Castro, L., Ochoa-Bojórquez, M.O., Sánchez-Tellez, J.L., Rojo-Cebreros, A.H. and Álvarez-Lajonchère, L. (2020). Advances in spotted rose snapper (*Lutjanus guttatus*, Steindachner, 1869) juveniles production. *Rev. Invest. Mar.*, 40 (2), xx-xx.